Search Notes 09/483,434 3/2/2004

L Number	Hits	Search Text	DB	Time stamp
1	0	WOJDA-$.IN. AND MILLER-J$.IN. AND GOLDSMITH-$.IN.	USPAT; US-PGPUB; EPO; JPO; DERWENT	2004/03/02 09:05
2	2	WOJDA-$.IN.	USPAT; US-PGPUB; EPO; JPO; DERWENT	2004/03/02 09:05
3	7597	WOJDA-$.IN. OR MILLER-J$.IN. OR GOLDSMITH-$.IN.	USPAT; US-PGPUB; EPO; JPO; DERWENT	2004/03/02 09:05
4	30	(WOJDA-$.IN. OR MILLER-J$.IN. OR GOLDSMITH-$.IN.) AND ((COVALENT$3 OR CONJUGAT$4 OR COMPLEX$4) WITH CELL)	USPAT; US-PGPUB; EPO; JPO; DERWENT	2004/03/02 09:05
5	0	((PEI OR POLYETHYLENIMINE) WITH AVIDIN) and (WOJDA-$.IN. OR MILLER-J$.IN. OR GOLDSMITH-$.IN.)	USPAT; US-PGPUB; EPO; JPO; DERWENT	2004/03/02 09:07
6	0	((PEI OR POLYETHYLENIMINE) WITH (avidin or streptavidin)) and sulfo-nhs-biotin	USPAT; US-PGPUB; EPO; JPO; DERWENT	2004/03/02 09:08
7	15	(PEI OR POLYETHYLENIMINE) WITH (avidin or streptavidin)	USPAT; US-PGPUB; EPO; JPO; DERWENT	2004/03/02 09:08
8	117	sulfo-nhs-lc-biotin	USPAT; US-PGPUB; EPO; JPO; DERWENT	2004/03/02 09:10
9	19	sulfo-nhs-lc--lc-biotin	USPAT; US-PGPUB; EPO; JPO; DERWENT	2004/03/02 09:10
10	268	sulfo-nhs-biotin or sulfo-nhs-lc-biotin or sulfo-nhs-lc-lc-biotin	USPAT; US-PGPUB; EPO; JPO; DERWENT	2004/03/02 09:10
11	201	(sulfo-nhs-biotin or sulfo-nhs-lc-biotin or sulfo-nhs-lc-lc-biotin) and (cell with surface)	USPAT; US-PGPUB; EPO; JPO; DERWENT	2004/03/02 09:10
12	48	(sulfo-nhs-biotin or sulfo-nhs-lc-biotin or sulfo-nhs-lc-lc-biotin) same (cell with surface)	USPAT; US-PGPUB; EPO; JPO; DERWENT	2004/03/02 09:10
13	11	(sulfo-nhs-biotin or sulfo-nhs-lc-biotin or sulfo-nhs-lc-lc-biotin) and 435/440-471.ccls.	USPAT; US-PGPUB; EPO; JPO; DERWENT	2004/03/02 09:11
14	16	(sulfo-nhs-biotin or sulfo-nhs-lc-biotin or sulfo-nhs-lc-lc-biotin) and (pei or polyethyleneimine)	USPAT; US-PGPUB; EPO; JPO; DERWENT	2004/03/02 09:11
15	47	((sulfo-nhs-biotin or sulfo-nhs-lc-biotin or sulfo-nhs-lc-lc-biotin) same (cell with (receptor or surface))) not (((sulfo-nhs-biotin or sulfo-nhs-lc-biotin or sulfo-nhs-lc-lc-biotin) and 435/440-471.ccls.) or ((sulfo-nhs-biotin or sulfo-nhs-lc-biotin or sulfo-nhs-lc-lc-biotin) and (pei or polyethyleneimine)))	USPAT; US-PGPUB; EPO; JPO; DERWENT	2004/03/02 09:11

16	34	(sulfo-nhs-biotin or sulfo-nhs-lc-biotin or sulfo-nhs-lc-lc-biotin) with (cell with surface)	USPAT; US-PGPUB; EPO; JPO; DERWENT	2004/03/02 09:11
18	16	(sulfo-nhs-biotin or sulfo-nhs-lc-biotin or sulfo-nhs-lc-lc-biotin) same (erythrocyte$1)	USPAT; US-PGPUB; EPO; JPO; DERWENT	2004/03/02 09:12
19	9	(sulfo-nhs-biotin or sulfo-nhs-lc-biotin or sulfo-nhs-lc-lc-biotin) same (transduc$6 or transfect$6)	USPAT; US-PGPUB; EPO; JPO; DERWENT	2004/03/02 09:12
20	1	(sulfo-nhs-biotin or sulfo-nhs-lc-biotin or sulfo-nhs-lc-lc-biotin) same ((gene or dna or polynucleotide or oligonucleotide or (nucleic adj2 acid) or plasmid or vector) with (deliver$4 or transform$7))	USPAT; US-PGPUB; EPO; JPO; DERWENT	2004/03/02 09:13
22	0	(sulfo-nhs-biotin or sulfo-nhs-lc-biotin or sulfo-nhs-lc-lc-biotin) same (tumor with cell$1)	USPAT; US-PGPUB; EPO; JPO; DERWENT	2004/03/02 09:13
23	0	(sulfo-nhs-biotin or sulfo-nhs-lc-biotin or sulfo-nhs-lc-lc-biotin) same (cancer$4 with cell$1)	USPAT; US-PGPUB; EPO; JPO; DERWENT	2004/03/02 09:13
24	15	(sulfo-nhs-biotin or sulfo-nhs-lc-biotin or sulfo-nhs-lc-lc-biotin) same (leukocyte$1)	USPAT; US-PGPUB; EPO; JPO; DERWENT	2004/03/02 09:14
26	0	((sulfo-nhs-biotin or sulfo-nhs-lc-biotin or sulfo-nhs-lc-lc-biotin) with cell$1).clm.	USPAT; US-PGPUB; EPO; JPO; DERWENT	2004/03/02 09:14
27	0	((sulfo-nhs-biotin or sulfo-nhs-lc-biotin or sulfo-nhs-lc-lc-biotin) with receptor).clm.	USPAT; US-PGPUB; EPO; JPO; DERWENT	2004/03/02 09:14
28	31	(biotin with cell with receptor).clm.	USPAT; US-PGPUB; EPO; JPO; DERWENT	2004/03/02 09:14
30	13	((sulfo-nhs-biotin or sulfo-nhs-lc-biotin or sulfo-nhs-lc-lc-biotin) and (pei or polyethyleneimine)) not ((sulfo-nhs-biotin or sulfo-nhs-lc-biotin or sulfo-nhs-lc-lc-biotin) and 435/440-471.ccls.)	USPAT; US-PGPUB; EPO; JPO; DERWENT	2004/03/02 09:15
31	20	(biotin$7) with cell with receptor with attach$4	USPAT; US-PGPUB; EPO; JPO; DERWENT	2004/03/02 09:15
32	20	(biotin$7) with cell with receptor with attach$4	USPAT; US-PGPUB; EPO; JPO; DERWENT	2004/03/02 09:15
33	4	((biotin$7) with cell with receptor with link$4).clm.	USPAT; US-PGPUB; EPO; JPO; DERWENT	2004/03/02 09:15
34	20	((biotin$7) with cell with receptor with attach$4) not (((biotin$7) with cell with receptor with link$4).clm.)	USPAT; US-PGPUB; EPO; JPO; DERWENT	2004/03/02 09:16

35	29	((biotin$7) with cell with receptor with link$4) not (((biotin$7) with cell with receptor with link$4).clm.)	USPAT; US-PGPUB; EPO; JPO; DERWENT	2004/03/02 09:16
25	1	(sulfo-nhs-biotin or sulfo-nhs-lc-biotin or sulfo-nhs-lc-lc-biotin) same (erythrocyte$1) and 6045795.pn.	USPAT; US-PGPUB; EPO; JPO; DERWENT	2004/03/02 09:16
17	9	(sulfo-nhs-biotin or sulfo-nhs-lc-biotin or sulfo-nhs-lc-lc-biotin) same (transduc$6 or transfect$6)	USPAT; US-PGPUB; EPO; JPO; DERWENT	2004/03/02 09:17
29	7	(sulfo-nhs-biotin or sulfo-nhs-lc-biotin or sulfo-nhs-lc-lc-biotin) same (lymphocyte$1)	USPAT; US-PGPUB; EPO; JPO; DERWENT	2004/03/02 09:20
21	3	(sulfo-nhs-biotin or sulfo-nhs-lc-biotin or sulfo-nhs-lc-lc-biotin) same (jurkat)	USPAT; US-PGPUB; EPO; JPO; DERWENT	2004/03/02 09:21

(FILE 'HOME' ENTERED AT 09:32:58 ON 02 MAR 2004)

FILE 'MEDLINE, CAPLUS, BIOSIS, EMBASE' ENTERED AT 09:33:21 ON 02 MAR 2004

L1	271771	S (WOJDA, ?)/IN,AU OR (MILLER, ?)/IN,AU OR (GOLDSMITH, ?)/IN,AU
L2	4	S (WOJDA, ?)/IN,AU AND (MILLER, ?)/IN,AU AND (GOLDSMITH, ?)/IN,
L3	2	DUPLICATE REMOVE L2 (2 DUPLICATES REMOVED)
L4	0	S L1 AND SULFO-NHS-LC-BIOTIN
L5	0	S L1 AND "SULFO-NHS-LC--LC-BIOTIN"
L6	1	S "SULFO-NHS-LC--LC-BIOTIN"
L7	32	S SULFO-NHS-LC-BIOTIN
L8	13	DUPLICATE REMOVE L7 (19 DUPLICATES REMOVED)
L9	11610	S POLYETHYLENIMINE OR PEI
L10	28	S L9 (S) (AVIDIN OR STREPTAVIDIN)
L11	36	S L9 (P) (AVIDIN OR STREPTAVIDIN)
L12	16	S L11 (P) (COVALENT? OR LINK? OR CROSSLINK?)
L13	6	DUPLICATE REMOVE L12 (10 DUPLICATES REMOVED)
L14	10	S L12 NOT L13
L15	20	S L11 NOT L12
L16	11	DUPLICATE REMOVE L15 (9 DUPLICATES REMOVED)

=>

ANSWER 8 OF 11 BIOSIS COPYRIGHT 2004 BIOLOGICAL ABSTRACTS INC. on STN

ACCESSION NUMBER: 1999:111564 BIOSIS

DOCUMENT NUMBER: PREV199900111564

TITLE: Gene delivery to biotinylated hematopoietic cells using **avidin-polyethylenimine** bioconjugates.

AUTHOR(S): Wojda, U.; Goldsmith, P.; Miller, J. L.

CORPORATE SOURCE: Lab. Chem. Biol. Metabol. Dis. Branch, Natl. Inst Diabetes Digestive Kidney Dis., NIH, Bethesda, MD, USA

SOURCE: Blood, (Nov. 15, 1998) Vol. 92, No. 10 SUPPL. 1 PART 1-2, pp. 382B. print.
Meeting Info.: 40th Annual Meeting of the American Society of Hematology. Miami Beach, Florida, USA. December 4-8, 1998. The American Society of Heamatology.
CODEN: BLOOAW. ISSN: 0006-4971.

DOCUMENT TYPE: Conference; (Meeting)
Conference; Abstract; (Meeting Abstract)

LANGUAGE: English

ENTRY DATE: Entered STN: 12 Mar 1999
Last Updated on STN: 12 Mar 1999

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* * * * * * * * * *   Welcome to STN International   * * * * * * * * * *

 NEWS  1           Web Page URLs for STN Seminar Schedule - N. America
 NEWS  2           "Ask CAS" for self-help around the clock
 NEWS  3  SEP 09   CA/CAplus records now contain indexing from 1907 to the
                   present
 NEWS  4  DEC 08   INPADOC: Legal Status data reloaded
 NEWS  5  SEP 29   DISSABS now available on STN
 NEWS  6  OCT 10   PCTFULL: Two new display fields added
 NEWS  7  OCT 21   BIOSIS file reloaded and enhanced
 NEWS  8  OCT 28   BIOSIS file segment of TOXCENTER reloaded and enhanced
 NEWS  9  NOV 24   MSDS-CCOHS file reloaded
 NEWS 10  DEC 08   CABA reloaded with left truncation
 NEWS 11  DEC 08   IMS file names changed
 NEWS 12  DEC 09   Experimental property data collected by CAS now available
                   in REGISTRY
 NEWS 13  DEC 09   STN Entry Date available for display in REGISTRY and CA/CAplus
 NEWS 14  DEC 17   DGENE: Two new display fields added
 NEWS 15  DEC 18   BIOTECHNO no longer updated
 NEWS 16  DEC 19   CROPU no longer updated; subscriber discount no longer
                   available
 NEWS 17  DEC 22   Additional INPI reactions and pre-1907 documents added to CAS
                   databases
 NEWS 18  DEC 22   IFIPAT/IFIUDB/IFICDB reloaded with new data and search fields
 NEWS 19  DEC 22   ABI-INFORM now available on STN
 NEWS 20  JAN 27   Source of Registration (SR) information in REGISTRY updated
                   and searchable
 NEWS 21  JAN 27   A new search aid, the Company Name Thesaurus, available in
                   CA/CAplus
 NEWS 22  FEB 05   German (DE) application and patent publication number format
                   changes

 NEWS EXPRESS  DECEMBER 28 CURRENT WINDOWS VERSION IS V7.00, CURRENT
               MACINTOSH VERSION IS V6.0b(ENG) AND V6.0Jb(JP),
               AND CURRENT DISCOVER FILE IS DATED 23 SEPTEMBER 2003
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FILE 'HOME' ENTERED AT 09:32:58 ON 02 MAR 2004

=> FILE MEDLINE CAPLUS BIOSIS EMBASE
COST IN U.S. DOLLARS                                  SINCE FILE      TOTAL
                                                         ENTRY    SESSION
FULL ESTIMATED COST                                       0.21       0.21

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=> S (WOJDA, ?)/IN,AU OR (MILLER, ?)/IN,AU OR (GOLDSMITH, ?)/IN,AU
'IN' IS NOT A VALID FIELD CODE
'IN' IS NOT A VALID FIELD CODE
L1 271771 (WOJDA, ?)/IN,AU OR (MILLER, ?)/IN,AU OR (GOLDSMITH, ?)/IN,AU

=> S (WOJDA, ?)/IN,AU AND (MILLER, ?)/IN,AU AND (GOLDSMITH, ?)/IN,AU
'IN' IS NOT A VALID FIELD CODE
'IN' IS NOT A VALID FIELD CODE
L2 4 (WOJDA, ?)/IN,AU AND (MILLER, ?)/IN,AU AND (GOLDSMITH, ?)/IN,AU

=> DUPLICATE REMOVE L2
DUPLICATE PREFERENCE IS 'MEDLINE, CAPLUS, BIOSIS'
KEEP DUPLICATES FROM MORE THAN ONE FILE? Y/(N):N
PROCESSING COMPLETED FOR L2
L3 2 DUPLICATE REMOVE L2 (2 DUPLICATES REMOVED)

=> D IBIB AB L3 1,2

L3 ANSWER 1 OF 2 MEDLINE on STN DUPLICATE 1
ACCESSION NUMBER: 2000031555 MEDLINE
DOCUMENT NUMBER: PubMed ID: 10563774
TITLE: Surface membrane biotinylation efficiently mediates the endocytosis of avidin bioconjugates into nucleated cells.
AUTHOR: **Wojda U; Goldsmith P; Miller J L**
CORPORATE SOURCE: Laboratory of Chemical Biology, National Institute of Diabetes and Digestive and Kidney Diseases and Metabolic Disease Branch, National Institutes of Health, Bethesda, Maryland 20892, USA.
SOURCE: Bioconjugate chemistry, (1999 Nov-Dec) 10 (6) 1044-50. Journal code: 9010319. ISSN: 1043-1802.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 200001
ENTRY DATE: Entered STN: 20000114
Last Updated on STN: 20000114
Entered Medline: 20000106

AB Here we demonstrate that biotin covalently attached to cell surface obligates existing receptors to endocytose avidin bioconjugates into nucleated cells. Incubation of fluorescein-labeled avidin with biotinylated cell lines resulted in uniform and rapid surface attachment and endocytosis compared with no detectable association of the avidin-conjugated dye with unbiotinylated cells. Uptake was detected within minutes with efficiencies approaching 100% in cell lines and freshly obtained peripheral blood mononuclear cells. After 24 h, avidin was barely detectable on the surface of the nucleated cells. In marked contrast, fluorescent avidin remained exclusively on the external membrane of erythrocytes after 24 h. To investigate biotin-mediated endocytosis for the delivery of DNA, we prepared polyethylenimine-avidin (PEI-avidin)

conjugates. Surface biotinylation significantly increased the transfection efficiencies of PEI-avidin condensed plasmid DNA coding green fluorescent protein (GFP) to the level of transferrin-receptor targeted gene delivery (15-20% GFP positive cells in culture after 48 h). The increase in transfection efficiency was blocked by the addition of free avidin or biotin to the culture medium. Biotin covalently bound to cell surface membrane proteins efficiently mediates the entry of avidin bioconjugates into nucleated cells.

L3 ANSWER 2 OF 2 BIOSIS COPYRIGHT 2004 BIOLOGICAL ABSTRACTS INC. on STN
ACCESSION NUMBER: 1999:111564 BIOSIS
DOCUMENT NUMBER: PREV199900111564
TITLE: Gene delivery to biotinylated hematopoietic cells using avidin-polyethylenimine bioconjugates.
AUTHOR(S): **Wojda, U.; Goldsmith, P.; Miller, J. L.**
CORPORATE SOURCE: Lab. Chem. Biol. Metabol. Dis. Branch, Natl. Inst Diabetes Digestive Kidney Dis., NIH, Bethesda, MD, USA
SOURCE: Blood, (Nov. 15, 1998) Vol. 92, No. 10 SUPPL. 1 PART 1-2, pp. 382B. print.
Meeting Info.: 40th Annual Meeting of the American Society of Hematology. Miami Beach, Florida, USA. December 4-8, 1998. The American Society of Heamatology.
CODEN: BLOOAW. ISSN: 0006-4971.
DOCUMENT TYPE: Conference; (Meeting)
Conference; Abstract; (Meeting Abstract)
LANGUAGE: English
ENTRY DATE: Entered STN: 12 Mar 1999
Last Updated on STN: 12 Mar 1999

=> D HIS

(FILE 'HOME' ENTERED AT 09:32:58 ON 02 MAR 2004)

FILE 'MEDLINE, CAPLUS, BIOSIS, EMBASE' ENTERED AT 09:33:21 ON 02 MAR 2004
L1 271771 S (WOJDA, ?)/IN,AU OR (MILLER, ?)/IN,AU OR (GOLDSMITH, ?)/IN,AU
L2 4 S (WOJDA, ?)/IN,AU AND (MILLER, ?)/IN,AU AND (GOLDSMITH, ?)/IN,
L3 2 DUPLICATE REMOVE L2 (2 DUPLICATES REMOVED)

=> S L1 AND SULFO-NHS-LC-BIOTIN
L4 0 L1 AND SULFO-NHS-LC-BIOTIN

=> S L1 AND "SULFO-NHS-LC--LC-BIOTIN"
L5 0 L1 AND "SULFO-NHS-LC--LC-BIOTIN"

=> S "SULFO-NHS-LC--LC-BIOTIN"
L6 1 "SULFO-NHS-LC--LC-BIOTIN"

=> S SULFO-NHS-LC-BIOTIN
L7 32 SULFO-NHS-LC-BIOTIN

=> D IBIB AB L6

L6 ANSWER 1 OF 1 CAPLUS COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 2002:531802 CAPLUS
DOCUMENT NUMBER: 137:243834
TITLE: In Vitro Selection for Catalytic Activity with Ribosome Display
AUTHOR(S): Amstutz, Patrick; Pelletier, Joelle N.; Guggisberg, Armin; Jermutus, Lutz; Cesaro-Tadic, Sandro; Zahnd, Christian; Plueckthun, Andreas

CORPORATE SOURCE: Biochemisches Institut, Universitaet Zuerich, Zurich, CH-8057, Switz.
SOURCE: Journal of the American Chemical Society (2002), 124(32), 9396-9403
CODEN: JACSAT; ISSN: 0002-7863
PUBLISHER: American Chemical Society
DOCUMENT TYPE: Journal
LANGUAGE: English
AB We report what is, to our knowledge, the first in vitro selection for catalytic activity based on catalytic turnover by using ribosome display, a method which does not involve living cells at any step. RTEM-β-lactamase was functionally displayed on ribosomes as a complex with its encoding mRNA. We designed and synthesized a mechanism-based inhibitor of β-lactamase, biotinylated ampicillin sulfone, appropriate for selection of catalytic activity of the ribosome-displayed β-lactamase. This derivative of ampicillin inactivated β-lactamase in a specific and irreversible manner. Under appropriate selection conditions, active RTEM-β-lactamase was enriched relative to an inactive point mutant over 100-fold per ribosome display selection cycle. Selection for binding, carried out with β-lactamase inhibitory protein (BLIP), gave results similar to selection with the suicide inhibitor, indicating that ribosome display is similarly efficient in catalytic activity and affinity selections. In the future, the capacity to select directly for enzymic activity using an entirely in vitro process may allow for a significant increase in the explorable sequence space relative to existing strategies.
REFERENCE COUNT: 53 THERE ARE 53 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

=> D HIS

(FILE 'HOME' ENTERED AT 09:32:58 ON 02 MAR 2004)

FILE 'MEDLINE, CAPLUS, BIOSIS, EMBASE' ENTERED AT 09:33:21 ON 02 MAR 2004
L1 271771 S (WOJDA, ?)/IN,AU OR (MILLER, ?)/IN,AU OR (GOLDSMITH, ?)/IN,AU
L2 4 S (WOJDA, ?)/IN,AU AND (MILLER, ?)/IN,AU AND (GOLDSMITH, ?)/IN,
L3 2 DUPLICATE REMOVE L2 (2 DUPLICATES REMOVED)
L4 0 S L1 AND SULFO-NHS-LC-BIOTIN
L5 0 S L1 AND "SULFO-NHS-LC--LC-BIOTIN"
L6 1 S "SULFO-NHS-LC--LC-BIOTIN"
L7 32 S SULFO-NHS-LC-BIOTIN

=> DUPLICATE REMOVE L7
DUPLICATE PREFERENCE IS 'MEDLINE, CAPLUS, BIOSIS, EMBASE'
KEEP DUPLICATES FROM MORE THAN ONE FILE? Y/(N):N
PROCESSING COMPLETED FOR L7
L8 13 DUPLICATE REMOVE L7 (19 DUPLICATES REMOVED)

=> D IBIB AB L8 1-13

L8 ANSWER 1 OF 13 MEDLINE on STN DUPLICATE 1
ACCESSION NUMBER: 2003298214 MEDLINE
DOCUMENT NUMBER: PubMed ID: 12825892
TITLE: Analysis of the envelope proteins of heat-shocked Vibrio parahaemolyticus cells by immunoblotting and biotin-labeling methods.
AUTHOR: Wong Hin-chung; Chen Yu-Chih
CORPORATE SOURCE: Department of Microbiology, Soochow University, Taipei, Taiwan 11102, Republic of China.. wonghc@mail.scu.edu.tw
SOURCE: Microbiology and immunology, (2003) 47 (5) 313-9.
Journal code: 7703966. ISSN: 0385-5600.

PUB. COUNTRY: Japan
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 200308
ENTRY DATE: Entered STN: 20030627
Last Updated on STN: 20030821
Entered Medline: 20030820

AB Vibrio parahaemolyticus, a common enteropathogen in tropical and subtropical coastal regions, exhibits significant adaptive acid tolerance response and heat-shock response, and the envelope proteins induced by stresses are suggested to be associated with virulence. This work examined the heat-shock proteins located in the envelope of V. parahaemolyticus by two rapid methods; namely, the immunoblotting and biotin-labeling methods. The bacterial cells were cultured at 25 C and heat shocked at 37 or 42 C for 1 or 2 hr. The cells were first lysed, then proteins were separated by gel electrophoresis and probed with antiserum raised against heat-shocked cells. Next, the heat-shocked cells were examined by labeling with water soluble **sulfo-NHS -LC-biotin**. Proteins of 33, 61, 66, 71, 78, 92 and 101 kDa were induced, while 55, 86, 102, 120 and 160 kDa proteins were markedly enhanced in the envelope of the heat-shocked V. parahaemolyticus cells. The biotin tagged envelope proteins were purified using a monomeric avidin column, and the N-terminal sequence was determined and compared with other high identity protein sequences. The sequence results suggest that Vph1 (55 kDa), Vph2 (46 kDa) and Vph3 (42 kDa) are de novo synthesized heat-shock proteins located in the envelope of this pathogen, and the functions of these proteins in stress protection and virulence have yet to be determined.

L8 ANSWER 2 OF 13 BIOSIS COPYRIGHT 2004 BIOLOGICAL ABSTRACTS INC. on STN
ACCESSION NUMBER: 2003:237737 BIOSIS
DOCUMENT NUMBER: PREV200300237737
TITLE: Non-radioactive method for labelling Trypanosoma brucei brucei (S427, clone 22) surface proteins using Ez-link **Sulfo-NHS-LC-Biotin** (Sulfosuccinimidyl-6- (Biotinamido) hexanoate).
AUTHOR(S): Ezeokonkwo, R. C. [Reprint Author]; Agu, W. E.; Black, S. J.
CORPORATE SOURCE: Dept. of Veterinary Parasitology and Entomology, University of Nigeria, Nsukka, Nigeria
SOURCE: Tropical Veterinarian, (2003) Vol. 21, No. 1, pp. 14-22. print.
ISSN: 0794-4845 (ISSN print).
DOCUMENT TYPE: Article
LANGUAGE: English
ENTRY DATE: Entered STN: 14 May 2003
Last Updated on STN: 14 May 2003

AB Here we describe a non-radioactive procedure that makes use of Ez-linkTM **Sulfo-NHS-LC-Biotin** Sulfosuccinimidyl-6-(Biotinamido) hexanoate to label the surface proteins of Trypanosoma brucei brucei (S427, clone 22). Different concentrations (100ug, 200ug, 400ug, 500ug) of Sulfosuccinimidyl-6-(biotinamido) hexanoate were used for the labelling at different temperatures of 4degreeC, room temperature, 37degreeC. The molecular weights of the labeled surface proteins were determined by running SDS-PAGE, and Western blot. Detection of the biotinylated surface proteins was done by the use of streptavidin-horseradish peroxidase conjugate and exposed with the ECL western blot detection reagent. It was observed that (i) the biological activities of the T. brucei brucei like the motility, viability, and growth were not affected adversely by the biotinylation; (ii) as little as 100ug **Sulfo-NHS-LC-Biotin** was

enough to label about 6X106 Trypanosoma brucei brucei; (iii) high temperature appears to have adverse effect on the viability and growth of biotinylated trypanosomas; (iv) three proteins of molecular weights 177.83, 141.25, and 79.43kDa whose actual identities and immunologic importance are not clear at this stage of the study were consistently and clearly labeled by the biotin at different concentrations at room temperature. In comparison with the other alternative methods of labelling surface proteins of parasites described in the literature, this method offers better advantages as there was no radioactive emission; no loss of biological activities like motility, viability, and growth at room temperature and 4degreeC. The method is less cumbersome, and requires less specialized equipment, and more importantly the method is very fast, stable, and reproducible.

L8 ANSWER 3 OF 13 BIOSIS COPYRIGHT 2004 BIOLOGICAL ABSTRACTS INC. on STN
ACCESSION NUMBER: 2003:543899 BIOSIS
DOCUMENT NUMBER: PREV200300539401
TITLE: RESTORATION OF BARRIER FUNCTION IN THE CORNEA.
AUTHOR(S): Hutcheon, A. E. K. [Reprint Author]; Sippel, K. C. [Reprint Author]; Zieske, J. D. [Reprint Author]
CORPORATE SOURCE: Schepens Eye Research Inst, Boston, MA, USA
SOURCE: ARVO Annual Meeting Abstract Search and Program Planner, (2003) Vol. 2003, pp. Abstract No. 3824. cd-rom. Meeting Info.: Annual Meeting of the Association for Research in Vision and Ophthalmology. Fort Lauderdale, FL, USA. May 04-08, 2003. Association for Research in Vision and Ophthalmology.
DOCUMENT TYPE: Conference; (Meeting)
Conference; Abstract; (Meeting Abstract)
LANGUAGE: English
ENTRY DATE: Entered STN: 19 Nov 2003
Last Updated on STN: 19 Nov 2003

AB Purpose: Previously we have studied the wound healing process by examining proliferation, growth factors and transcription factors in both the corneal epithelium and stroma; these studies have indicated that wound healing in the cornea is a complex process with multiple interactions. Our current study examines the possibility of a correlation between the restoration of barrier function and basement membrane reassembly in the cornea after wounding. Methods: Adult Sprague-Dawley rats were anesthetized and a 3mm superficial keratectomy was performed. The eyes were allowed to heal from 4 hours to 8 weeks. Four rats for each time point had the right eye wounded, and the contralateral eye served as a control. At the time of sacrifice, EZ-Link **Sulfo-NHS-LC-Biotin** (Pierce; Rockford, IL) was applied to all eyes, experimental and control, for 15 minutes. This compound does not penetrate through intact tight junctions. After 15 minutes, the eyes were rinsed with 1x PBS, blotted, enucleated and frozen in OCT on dry ice. Six mum sections were cut and indirect immunofluorescence was performed with anti-laminin (Dako; Carpenteria, CA), a marker of basement membrane, and anti-streptavidin (Jackson ImmunoResearch; West Grove, PA), a marker of biotin. The tissue was rehydrated, blocked and incubated for 1 hour at room temperature with anti-laminin. After an hour, the tissue was rinsed, blocked and incubated with anti-rabbit IgG and streptavidin-TRITC for another hour at room temperature. Tissues were then rinsed and coverslipped with Vectashield mounting media containing DAPI (Vector Labs; Burlingame, CA), a marker of nuclei. Results: Upon wounding, biotin appears to penetrate into the stroma subjacent and slightly peripheral to the wound area. This pattern is present from 4-32 hours post-wounding; however, as the cornea heals, the basement membrane reassembles, as shown by laminin localization. The area of biotin localization appears to decrease at these time points. By 48 hours, the basement membrane has fully reassembled throughout the entire wound area and no biotin is

present in the stroma. Conclusions: Reformation of the basement membrane appears to correlate with the restoration of the barrier function in the cornea.

L8 ANSWER 4 OF 13 BIOSIS COPYRIGHT 2004 BIOLOGICAL ABSTRACTS INC. on STN
ACCESSION NUMBER: 2003:270699 BIOSIS
DOCUMENT NUMBER: PREV200300270699
TITLE: Investigating the stability of the biotin-protein bond in human plasma.
AUTHOR(S): Mock, Donald M. [Reprint Author]; Mock, Nell I.; Bogusiewicz, Anna
CORPORATE SOURCE: Biochemistry and Molecular Biology, Univeristy of Arkansas for Medical Sciences, 4301 West Markham, Little Rock, AR, 72205, USA
mockdonaldm@uams.edu; mocknelli@uams.edu; bogusiewiczanna@uams.edu
SOURCE: FASEB Journal, (March 2003) Vol. 17, No. 4-5, pp. Abstract No. 439.19. http://www.fasebj.org/. e-file.
Meeting Info.: FASEB Meeting on Experimental Biology: Translating the Genome. San Diego, CA, USA. April 11-15, 2003. FASEB.
ISSN: 0892-6638 (ISSN print).
DOCUMENT TYPE: Conference; (Meeting)
Conference; Abstract; (Meeting Abstract)
LANGUAGE: English
ENTRY DATE: Entered STN: 11 Jun 2003
Last Updated on STN: 11 Jun 2003

AB Biotinylated proteins offer a safe alternative to radioactive tracers in human pharmacokinetic studies. Stability of the biotin-protein bond in human plasma is crucial. We examined the stability of the biotin-protein bond using IgG biotinylated with five agents: biotin PEO-amine (BPEO), 5-(biotinamido)-pentylamine (5BP), iodoacetyl-LC-biotin (IAB), **sulfo-NHS-LC-biotin** (NHSB), and biotin-LC-hydrazide (BH). Stability of the biotin bond was quantitated by directly measuring biotin release. Each biotinylated protein was incubated at room temperature for 4h in either human plasma or phosphate-buffered saline (PBS) + 0.02% Tween 20. Free biotin was separated from the biotinylated protein by ultrafiltration using 10,000 dalton exclusion filters; the biotin concentration in the ultrafiltrate was then quantitated by avidin-binding assay. Biotin release rate from total bound biotin %Biotinylation reagentplasma PBS bufferBPEO8.2+-50.3+-65BP6.5+-30.3+-4NHSB18+-70.3+-5IAB12.2+-60.2+-9BH13.6+-30.6+-4Stability of the biotin-protein amide bond formed by NHSB was also assessed by quantitating capture of europium streptavidin (Eu-SA) by the biotinylated IgG immobilized on a protein A-coated microtiter well. Eu-SA binding was reduced by 91.5% after incubation for 4h with plasma compared to 4h buffer control. We conclude that the 5 biotinylating agents produced biotin-protein bonds that were stable in PBS but susceptible to hydrolysis by factors present in human plasma.

L8 ANSWER 5 OF 13 BIOSIS COPYRIGHT 2004 BIOLOGICAL ABSTRACTS INC. on STN
ACCESSION NUMBER: 2002:369433 BIOSIS
DOCUMENT NUMBER: PREV200200369433
TITLE: Tryptophan and serine residues may be involved in the function of the human Na+/sulfate cotransporter, hNaSi-1.
AUTHOR(S): Li, Hongyan [Reprint author]; Pajor, Ana M. [Reprint author]
CORPORATE SOURCE: Physiology and Biophysics, Medical Branch, University of Texas, 301 University Blvd. Route 0641, Galveston, TX, 77555, USA
SOURCE: FASEB Journal, (March 22, 2002) Vol. 16, No. 5, pp. A807. print.

Meeting Info.: Annual Meeting of Professional Research Scientists on Experimental Biology. New Orleans, Louisiana, USA. April 20-24, 2002.
CODEN: FAJOEC. ISSN: 0892-6638.

DOCUMENT TYPE: Conference; (Meeting)
Conference; Abstract; (Meeting Abstract)
LANGUAGE: English
ENTRY DATE: Entered STN: 3 Jul 2002
Last Updated on STN: 3 Jul 2002

AB The Na+/sulfate cotransporter, NaSi-1, is located on the apical membrane of the renal proximal tubular epithelial cells, where it acts to reabsorb filtered sulfate. NaSi-1 belongs to a distinct gene family, SLC13, which also includes the Na+/dicarboxylate cotransporters. The hNaSi-1 was amplified from human kidney cDNA using specific primers based on Genbank AK026413 and the function was verified by transport measurements in Xenopus oocytes. We mutated some of the conserved tryptophan, serine, and aspartic acid residues in hNaSi-1 to identify potential substrate binding sites based on previous studies that these residues were involved in sulfate binding to the bacterial sulfate transport receptor. The expression of the proteins in oocytes was detected using cell-surface biotinylation with **Sulfo-NHS-LC-Biotin** and Western blots with antibodies raised against a fusion protein of hNaSi-1 and GST. All of the mutations in which tryptophan was replaced by alanine resulted in transporters with little or no transport activity, although the proteins were expressed at the cell surface. Mutations of conserved serines resulted in about 50%-80% reduction of activity, whereas mutations of aspartic acid residues had little effect on transport. We conclude that tryptophan and serine residues have functional roles in hNaSi-1 and experiments are currently underway to determine in more detail how these residues act in substrate binding or translocation.

L8 ANSWER 6 OF 13 BIOSIS COPYRIGHT 2004 BIOLOGICAL ABSTRACTS INC. on STN
ACCESSION NUMBER: 2003:154579 BIOSIS
DOCUMENT NUMBER: PREV200300154579
TITLE: Isolated Muramyl Dipeptide Receptor Is Identified as A Special Cell Surface Calreticulin.
AUTHOR(S): Chen, D. [Reprint Author]; Langford, M. P.; Duggan, C. [Reprint Author]; Ganley, J. P. [Reprint Author]; Texada, D. E. [Reprint Author]
CORPORATE SOURCE: Ophthalmology, LSU Health Sciences Center, Shreveport, LA, USA
SOURCE: ARVO Annual Meeting Abstract Search and Program Planner, (2002) Vol. 2002, pp. Abstract No. 2409. cd-rom.
Meeting Info.: Annual Meeting of the Association For Research in Vision and Ophthalmology. Fort Lauderdale, Florida, USA. May 05-10, 2002.
DOCUMENT TYPE: Conference; (Meeting)
Conference; Abstract; (Meeting Abstract)
LANGUAGE: English
ENTRY DATE: Entered STN: 26 Mar 2003
Last Updated on STN: 26 Mar 2003

AB We have found that the natural bacterial cell wall component muramyl dipeptide (MDP) can induce apoptotic DNA ladders in rabbit tears. And it can also induce primary cultured rabbit retinal cells and rabbit kidney cell line RK13 cells to go apoptotic in a dose-dependent manner. Moreover, we have already determined that the receptor mediating the MDP-induced apoptosis of RK13 cells is on its cell surface. Purpose: To isolate and identify the MDP receptor from RK13 cells. Methods: Sepharose 4B coupled MDP (MDP-SP4B) was first synthesized. The MDP binding protein (MBP) of RK13 cells was isolated by MDP-SP4B affinity chromatography. Isolated MBP was analyzed by mass spectrometry, and spectra data were

analyzed by Auto MS-Fit Software (UCSF) against NCBI and Genpept protein databases (protein identification analysis). Moreover, RK13 cells were surface labeled by **Sulfo-NHS-LC-Biotin** and calreticulin (CRT) was then purified from the biotinylated cells by mannan-agarose affinity chromatography. Western blotting analysis of purified CRT was performed with both horseradish peroxidase conjugated streptavidin (S-HRP) and anti-CRT antibody/horseradish peroxidase conjugated secondary antibody. Free MDP binding activity of both isolated MBP and purified CRT was determined by apoptosis inhibition method. Results: The molecular weight of the isolated MBP is 56 kilodalton (kD) and it had free MDP binding activity. Protein identification analysis suggested MBP is calreticulin. ELISA first confirmed MBP was calreticulin. Western blotting analysis showed purified CRT had 2 protein bands: 56 kD and 60 kD. And only 56 kD band could be detected by S-HRP. Eluate which just contained 56 kD band could exhibit strong free MDP binding activity while eluate with both bands showed very weak free MDP binding activity. Conclusions: Our isolated MDP binding protein is a special kind of calreticulin with molecular weight of 56 kD, which resides on the RK13 cell surface and can mediate MDP-induced apoptosis.

L8 ANSWER 7 OF 13 MEDLINE on STN DUPLICATE 2
ACCESSION NUMBER: 2001518507 MEDLINE
DOCUMENT NUMBER: PubMed ID: 11565800
TITLE: Differential extraction and enrichment of human sperm surface proteins in a proteome: identification of immunocontraceptive candidates.
AUTHOR: Shetty J; Diekman A B; Jayes F C; Sherman N E; Naaby-Hansen S; Flickinger C J; Herr J C
CORPORATE SOURCE: Department of Cell Biology, University of Virginia, Charlottesville 22908-0732, USA.
CONTRACT NUMBER: D43 TW/HD 00654 (FIC)
HD U54 29099 (NICHD)
SOURCE: Electrophoresis, (2001 Aug) 22 (14) 3053-66.
Journal code: 8204476. ISSN: 0173-0835.
PUB. COUNTRY: Germany: Germany, Federal Republic of
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 200204
ENTRY DATE: Entered STN: 20010924
Last Updated on STN: 20020409
Entered Medline: 20020408

AB The objective of this study was to discover previously unknown human sperm surface proteins that may be candidate contraceptive vaccinogens. To this end, methods of concentrating human sperm proteins for microsequencing by mass spectrometry were used, which increased the likelihood of identifying surface proteins. Vectorial labeling, differential extraction and two-dimensional (2-D) gel electrophoresis were employed to identify and isolate proteins accessible at the cell surface. Percoll harvested or swim-up sperm were either solubilized directly or solubilized after surface labeling with sulfo-succinimidyl-6-(biotinamido)hexanoate (**sulfo-NHS-LC-biotin**). Comparisons were made of proteins extracted with four lysis buffers: (i) Celis buffer containing 9.8 M urea and 2% Igepal CA-630; (ii) 1% Triton X (TX)-100; (iii) 1.7% TX-114 followed by phase partitioning; or (iv) 1 M NaCl. Blots of proteins separated by high-resolution 2-D electrophoresis were probed with avidin and antibodies to known proteins specific for three domains: the sperm surface (SAGA-1), the acrosome (SP-10), and the cytoskeleton (alpha-tubulin). Celis buffer (45 min) extracted proteins from all three major compartments. However, a 20-s extraction in Celis buffer enriched for several proteins and enabled the identification of several novel

peptides by mass spectrometry. Mild extraction with TX-100 or 1 M NaCl solubilized mainly membrane and acrosomal proteins, but not cytoskeletal proteins. Comparison of biotinylated proteins extracted by each method showed that the major vectorially labeled proteins solubilized by Celis buffer were also solubilized by TX-100, TX-114, and 1 M NaCl. Extraction with TX-114 followed by phase-partitioning significantly enriched hydrophobic surface proteins and aided resolution and isolation. Eight protein spots microsequenced following all these extraction methods proved to be novel sperm molecules.

L8 ANSWER 8 OF 13 MEDLINE on STN DUPLICATE 3
ACCESSION NUMBER: 2002031691 MEDLINE
DOCUMENT NUMBER: PubMed ID: 11757779
TITLE: A novel intravascular drug delivery method using endothelial biotinylation and avidin-biotin binding.
AUTHOR: Hoya K; Guterman L R; Miskolczi L; Hopkins L N
CORPORATE SOURCE: Department of Neurosurgery, School of Medicine and Biomedical Sciences, State University of New York at Buffalo, USA.. khoya@dokkyomed.ac.jp
SOURCE: Drug delivery, (2001 Oct-Dec) 8 (4) 215-22. Journal code: 9417471. ISSN: 1071-7544.
PUB. COUNTRY: England: United Kingdom
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 200203
ENTRY DATE: Entered STN: 20020124
Last Updated on STN: 20020307
Entered Medline: 20020306

AB In this study, a novel intravascular drug delivery system was developed in which a drug injected from a catheter was fixed to the vasculature of the targeted tissue. Cellular proteins of viable endothelial cells were first biotinylated directly by biotinylation reagents, and then bound by an avidinated drug or, using avidin as a linker, a biotinylated drug. In the initial experiments, we studied in vitro the biotinylation of cultured bovine aortic endothelial cells (BAECs) by applying biotinylation reagents (NHS-LC-biotin or **sulfo-NHS-LC-biotin**) onto the washed intact BAEC monolayers and showed that the amount of biotin bound to the cells depended on the concentration of the biotinylation reagents applied. The cell-bound biotin decreased with time after the biotinylation. When fluorescein-labeled avidin (FITC-avidin) was applied to the biotinylated BAEC monolayers, the FITC-avidin readily bound to the cells. An LDH-release assay showed that **sulfo-NHS-LC-biotin** was only slightly cytotoxic to the BAECs and a colony formation assay showed only slight adverse effects of the reagent. In vivo studies were carried out on the renal arteries of normal rabbits. A solution of NHS-LC-biotin was injected through a catheter to one kidney to biotinylate its vasculature and the vehicle to the other as control, followed by a perfusion with saline. Finally, a solution of FITC-avidin was injected to both kidneys that were then reperfused with the blood flow following the withdrawal of the catheters. In the histological sections, more than 85% of glomeruli was stained with fluorescein in the biotinylated kidney, whereas no glomeruli were stained in the control. In the kidneys harvested 2 days after the same procedure, most glomeruli were still brightly stained. In the final experiment, biotinylated kidneys were injected with a solution of avidin, followed by a solution of fluorescein-biotin. Control kidneys had no prior biotinylation but received the same injections of avidin and fluorescein-biotin as above. More than 80% of glomeruli were stained in the biotinylated kidneys but none in the controls. This indicated that biotinylated drugs can be anchored to the biotinylated vasculature through avidin without being flushed away by blood flows. No apparent adverse

effect was found in the functions of biotinylated kidneys. We propose that this drug delivery system is feasible for the treatment of some pathological conditions of blood vessels such as microvascular proliferation in malignant tumors and for continuous drug delivery in certain target organs.

L8 ANSWER 9 OF 13 MEDLINE on STN DUPLICATE 4
ACCESSION NUMBER: 1998422453 MEDLINE
DOCUMENT NUMBER: PubMed ID: 9748433
TITLE: New potential cell wall glucanases of Saccharomyces cerevisiae and their involvement in mating.
AUTHOR: Cappellaro C; Mrsa V; Tanner W
CORPORATE SOURCE: Lehrstuhl fur Zellbiologie und Pflanzenphysiologie, Universitat Regensburg, 93040 Regensburg, Germany.
SOURCE: Journal of bacteriology, (1998 Oct) 180 (19) 5030-7. Journal code: 2985120R. ISSN: 0021-9193.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199810
ENTRY DATE: Entered STN: 19981029
Last Updated on STN: 19981029
Entered Medline: 19981019

AB Biotinylation of intact Saccharomyces cerevisiae cells with a nonpermeant reagent (**Sulfo-NHS-LC-Biotin**) allowed the identification of seven cell wall proteins that were released from intact cells by dithiothreitol (DTT). By N-terminal sequencing, three of these proteins were identified as the known proteins beta-exoglucanase 1 (Exg1p), beta-endoglucanase (Bgl2p), and chitinase (Cts1p). One protein was related to the PIR protein family, whereas the remaining three (Scw3p, Scw4p, and Scw10p [for soluble cell wall proteins]) were found to be related to glucanases. Single knockouts of these three potential glucanases did not result in dramatic phenotypes. The double knockout of SCW4 and the homologous gene SCW10 resulted in slower growth, significantly increased release of proteins from intact cells by DTT, and highly decreased mating efficiency when these two genes were disrupted in both mating types. The synergistic behavior of the disruption of SCW4 and SCW10 was partly antagonized by the disruption of BGL2. The data are discussed in terms of a possible counterplay of transglucosidase and glucosidase activities.

L8 ANSWER 10 OF 13 MEDLINE on STN DUPLICATE 5
ACCESSION NUMBER: 1998450583 MEDLINE
DOCUMENT NUMBER: PubMed ID: 9777408
TITLE: Structure and function of gC1q-R: a multiligand binding cellular protein.
AUTHOR: Ghebrehiwet B; Peerschke E I
CORPORATE SOURCE: Department of Medicine, State University of New York, Stony Brook, USA.. berhane@mail.som.sunysb.edu
CONTRACT NUMBER: R01HL5029101 (NHLBI)
SOURCE: Immunobiology, (1998 Aug) 199 (2) 225-38. Ref: 58
Journal code: 8002742. ISSN: 0171-2985.
PUB. COUNTRY: GERMANY: Germany, Federal Republic of
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
General Review; (REVIEW)
(REVIEW, TUTORIAL)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199901
ENTRY DATE: Entered STN: 19990115
Last Updated on STN: 19990115

Entered Medline: 19990107

AB gC1q-R is a 33 kDa, single chain, highly acidic protein, which was first isolated from membrane preparation of Raji cells and now appears to be ubiquitously distributed. Although, gC1q-R was originally identified as a protein which binds to the globular "heads" of C1q, recent evidence suggests that the molecule is in fact a multiligand binding, multifunctional protein with affinity for diverse ligands which at best are functionally related. These molecules include: thrombin, vitronectin, and high molecular weight kininogen. The gC1q-R molecule, which is identical to the transcription factors SF2 and the Tat-associated protein, or TAP, is the product of a single gene localized on chromosome 17p13.3 in human, and chromosome 11 in mouse, and is encoded by an approximately 1.5-1.6 kb mRNA. The full length cDNA encodes a primary translation protein of 282 residues and the 'mature' or membrane form of the protein isolated from Raji cells corresponds to residues 74-282 and is presumed to be generated by a site-specific cleavage and removal of the highly basic, 73-residues long, N-terminal segment during post-translational processing. The translated amino acid sequence does not predict for the presence of a conventional sequence motif compatible with a transmembrane segment and does not have a consensus site for a GPI anchor. However, there is strong evidence which indicates that gC1q-R is expressed both inside the cell and on the membrane. First, certain mAbs raised against gC1q-R react moderately with intact Raji cells in suspension and this binding increases when the cells are first bound to poly-L-lysine coated surfaces and then fixed with glutaraldehyde. Second, surface labeling of cells using the membrane impermeable **sulfo-NHS-LC-biotin** shows that gC1q-R on the surface incorporates biotin whereas intracellular gC1q-R does not. In addition, the membrane expression of gC1q-R can be upregulated with inflammatory cytokines such as INF-gamma, TNF-alpha, or LPS. These results suggest, that gC1q-R, is localized both as an intracellular and as a cell surface protein and may have important biological functions in both compartments of the cell.

L8 ANSWER 11 OF 13 BIOSIS COPYRIGHT 2004 BIOLOGICAL ABSTRACTS INC. on STN

ACCESSION NUMBER: 1991:409274 BIOSIS
DOCUMENT NUMBER: PREV199192076239; BA92:76239
TITLE: SULFO-N-HYDROXYSUCCINIMIDE ACTIVATED LONG CHAIN BIOTIN A NEW MICROTITER PLATE ASSAY FOR THE DETERMINATION OF ITS STABILITY AT DIFFERENT PH VALUES AND ITS REACTION RATE WITH PROTEIN BOUND AMINO GROUPS.
AUTHOR(S): GRUMBACH I M [Reprint author]; VEH R W
CORPORATE SOURCE: ABT NEUROANATOMONIE, RUHR-UNIV BOCHUM UNIV, 150, D-4630 BOCHUM, GER
SOURCE: Journal of Immunological Methods, (1991) Vol. 140, No. 2, pp. 205-210.
CODEN: JIMMBG. ISSN: 0022-1759.
DOCUMENT TYPE: Article
FILE SEGMENT: BA
LANGUAGE: ENGLISH
ENTRY DATE: Entered STN: 11 Sep 1991
Last Updated on STN: 11 Sep 1991

AB Biotinamidohexanoic acid N-hydroxysulfosuccinimide ester (N-hydroxysulfosuccinimide activated long chain biotin; **sulfo-NHS-LC-biotin**) has become an invaluable tool for the biotinylation of protein despite the absence of data concerning its stability and reaction velocity. A convenient, rapid and sensitive assay for this compound has been developed based on the **sulfo-NHS-LC biotin** mediated biotinylated of bovine serum albumin following adsorption to the wells of a microtiter plate. Bound biotin was visualized by the sequential use of streptavidin and biotinylated horseradish peroxidase. This assay was used for the determination of the stability of **sulfo-NHS-LC**

-biotin in aqueous solution of different pH values. Hydrolysis half lives were below 15 min at pH values above 8.0, but at pH values below 6.5 they exceeded 2 h. It is suggested, therefore, that biotinylations should be performed with **sulfo-NHS-LC-biotin** taken from a stock solution, prepared at pH values between 3.0 and 5.8. Reaction velocities with primary amino groups were also investigated by means of this ELISA procedure. As expected, biotinylated proceeds faster at pH 8.0 as compared to 7.2, but the increased reaction rate does not compensate for the decreased hydrolysis half life at the higher pH value. Thus, biotinylation with **sulfo-NHS-LC-biotin** at near neutral pH values appears to be optimal.

L8 ANSWER 12 OF 13 MEDLINE on STN DUPLICATE 6
ACCESSION NUMBER: 90357783 MEDLINE
DOCUMENT NUMBER: PubMed ID: 2389556
TITLE: Reovirus binds to multiple plasma membrane proteins of mouse L fibroblasts.
AUTHOR: Choi A H; Paul R W; Lee P W
CORPORATE SOURCE: Department of Microbiology and Infectious Diseases, University of Calgary Health Sciences Centre, Alberta, Canada.
SOURCE: Virology, (1990 Sep) 178 (1) 316-20.
Journal code: 0110674. ISSN: 0042-6822.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199009
ENTRY DATE: Entered STN: 19901026
Last Updated on STN: 19970203
Entered Medline: 19900924

AB Plasma membranes from mouse L fibroblasts were isolated and subjected to sodium dodecyl sulfate-polyacrylamide gel electrophoresis (SDS-PAGE). Resolved proteins were electroblotted to nitrocellulose paper and probed with 125I-labeled type 3 (T3) reovirus. Multiple protein bands with molecular weights ranging from 26 to 200 kDa were consistently recognized by the virus. Such binding was specific since it was blocked in the presence of unlabeled virus. That these proteins were exposed on the cell surface was confirmed by their susceptibility to **sulfo-NHS-LC-biotin** labeling of intact cells prior to membrane purification. Blots probed with wheat germ agglutinin (WGA)-gold showed a similar pattern of protein bands. These findings are consistent with the ability of WGA to block reovirus binding to L cells, and with our recent demonstration that the alpha-anomeric form of sialic acid is the minimal receptor determinant recognized by reovirus (R. W. Paul, A. H. C. Choi, and P. W. K. Lee, Virology 172, 382-385, 1989). Both type 1 and type 3 reoviruses were found to recognize the same set of multiple proteins on the blot, which is again consistent with the previous observation that the two serotypes compete with each other for binding to intact L cells.

L8 ANSWER 13 OF 13 MEDLINE on STN DUPLICATE 7
ACCESSION NUMBER: 90036829 MEDLINE
DOCUMENT NUMBER: PubMed ID: 2808341
TITLE: Biotinylation of reactive amino groups in native recombinant human interleukin-1 beta.
AUTHOR: Yem A W; Zurcher-Neely H A; Richard K A; Staite N D; Heinrikson R L; Deibel M R Jr
CORPORATE SOURCE: Department of Biopolymer Chemistry, Upjohn Company, Kalamazoo, Michigan 49001.
SOURCE: Journal of biological chemistry, (1989 Oct 25) 264 (30)

17691-7.
Journal code: 2985121R. ISSN: 0021-9258.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 198911
ENTRY DATE: Entered STN: 19900328
Last Updated on STN: 19900328
Entered Medline: 19891127

AB Recombinant human interleukin-1 beta (rIL-1 beta) was chemically modified by a 10-fold molar excess (reagent:protein) of sulfosuccinimidyl 6-(biotinamido) hexanoate (**sulfo-NHS-LC-biotin**) or sulfosuccinimidobiotin (sulfo-NHS-biotin) under mild conditions. The primary product was purified in each case by cation exchange high performance liquid chromatography (HPLC) and digested with endoproteinase Lys C. Peptide mapping by C18 reverse phase HPLC permitted identification of three sites of biotinylation using both reagents; N-terminal alanine, lysine 93, and lysine 94. Few additional singly modified rIL-1 beta products were obtained under these conditions, despite the presence of 15 lysine residues in this protein. These data support the view that the N terminus as well as the trilysine sequence (residues 92-94) are readily susceptible to chemical modification and are exposed on the surface of the protein. Chromatography of intact biotinylated rIL-1 beta by C4 reverse phase HPLC resolved a protein modified exclusively at the N-terminal alanine from two proteins modified singly at either lysine 93 or lysine 94. In addition, a protein product modified at lysine 103 was also obtained when rIL-1 beta was similarly modified with sulfo-NHS-biotin. Since the only difference between the two biotinylation reagents relates to spacer length and its associated hydrophobicity, these data suggest that lysine 103 is not as accessible to surface modification reagents as are lysine 93, lysine 94, or alanine 1. Initial experiments indicate that none of the modifications described above decrease thymocyte proliferation by more than one order of magnitude. Therefore, these amino acid residues are not crucial for bioactivity, and we anticipate the use of these monobiotinylated proteins in structure/function analysis of IL-1 beta.

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=> D HIS

     (FILE 'HOME' ENTERED AT 09:32:58 ON 02 MAR 2004)

     FILE 'MEDLINE, CAPLUS, BIOSIS, EMBASE' ENTERED AT 09:33:21 ON 02 MAR 2004
L1        271771 S (WOJDA, ?)/IN,AU OR (MILLER, ?)/IN,AU OR (GOLDSMITH, ?)/IN,AU
L2             4 S (WOJDA, ?)/IN,AU AND (MILLER, ?)/IN,AU AND (GOLDSMITH, ?)/IN,
L3             2 DUPLICATE REMOVE L2 (2 DUPLICATES REMOVED)
L4             0 S L1 AND SULFO-NHS-LC-BIOTIN
L5             0 S L1 AND "SULFO-NHS-LC--LC-BIOTIN"
L6             1 S  "SULFO-NHS-LC--LC-BIOTIN"
L7            32 S SULFO-NHS-LC-BIOTIN
L8            13 DUPLICATE REMOVE L7 (19 DUPLICATES REMOVED)

=> S POLYETHYLENIMINE OR PEI
L9         11610 POLYETHYLENIMINE OR PEI

=> S L9 (S) (AVIDIN OR STREPTAVIDIN)
L10           28 L9 (S) (AVIDIN OR STREPTAVIDIN)

=> S L9 (P) (AVIDIN OR STREPTAVIDIN)
L11           36 L9 (P) (AVIDIN OR STREPTAVIDIN)
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=> S L11 (P) (COVALENT? OR LINK? OR CROSSLINK?)
L12 16 L11 (P) (COVALENT? OR LINK? OR CROSSLINK?)

=> DUPLICATE REMOVE L12
DUPLICATE PREFERENCE IS 'MEDLINE, CAPLUS, BIOSIS, EMBASE'
KEEP DUPLICATES FROM MORE THAN ONE FILE? Y/(N):N
PROCESSING COMPLETED FOR L12
L13 6 DUPLICATE REMOVE L12 (10 DUPLICATES REMOVED)

=> D IBIB AB L13 1-6

L13 ANSWER 1 OF 6 CAPLUS COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 2003:579142 CAPLUS
TITLE: One hundred spots parallel monitoring of DNA interactions by SPR imaging of polymer-functionalized surfaces applied to the detection of cystic fibrosis mutations
AUTHOR(S): Bassil, Nathalie; Maillart, Emmanuel; Canva, Michael; Levy, Yves; Millot, Marie-Claude; Pissard, Serge; Narwa, Remy; Goossens, Michel
CORPORATE SOURCE: Centre National de la Recherche Scientifique CNRS UMR 8501, Laboratoire Charles Fabry de l'Institut d'Optique (LCFIO), Universite Paris XI, Orsay, 91403, Fr.
SOURCE: Sensors and Actuators, B: Chemical (2003), B94(3), 313-323
CODEN: SABCEB; ISSN: 0925-4005
PUBLISHER: Elsevier Science B.V.
DOCUMENT TYPE: Journal
LANGUAGE: English
AB In the present paper, we report the detection of mutations implicated in human cystic fibrosis (CF). Nine different oligonucleotides are studied, including three possible mutations related to this specific genetic disease: a deletion of three bases, ΔF508, and two single-nucleotide polymorphisms 1540A/G and 1716G/A. We monitor, in real time and in parallel, hybridizations of a solution of unlabeled oligonucleotide targets to a matrix of 100 spots of oligonucleotide probes using surface plasmon resonance (SPR) imaging of a bio-functionalized gold slide. In order to functionalize our gold slide with the DNA probes, we have developed a self-assembled multilayer (SAM) based on electrostatic interactions and formed with 11-mercaptoundecanoic acid (MUA), poly(ethylenimine) (**PEI**) and ExtrAvidin layers. Probes are then **linked** to this SAM by the usual strong binding affinity of the **avidin** -biotin duplex. The 100 spots array deposited by a robot can be addressed either several times, sequentially, with the various oligonucleotide targets, or once, in parallel, with a mixture of some oligonucleotides. The specific response of our system is established along with the possibility of discriminating between a totally complementary sequence and its mutant form, even for a single base mismatch thus demonstrating the capacity of parallel diagnostic using patient like material.
REFERENCE COUNT: 28 THERE ARE 28 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L13 ANSWER 2 OF 6 MEDLINE on STN DUPLICATE 1
ACCESSION NUMBER: 2000469399 MEDLINE
DOCUMENT NUMBER: PubMed ID: 10930517
TITLE: Glycosylphosphatidylinositol-anchored proteins are not required for crosslinking-mediated endocytosis or transfection of avidin bioconjugates into biotinylated cells.
AUTHOR: Wojda U; Miller J L
CORPORATE SOURCE: Laboratory of Chemical Biology, National Institute of

Diabetes and Digestive and Kidney Diseases, National Institutes of Health, Bethesda, MD 20892, USA.

SOURCE: Biochimica et biophysica acta, (2000 Jul 31) 1467 (1) 144-52.
Journal code: 0217513. ISSN: 0006-3002.

PUB. COUNTRY: Netherlands
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 200010
ENTRY DATE: Entered STN: 20001012
Last Updated on STN: 20001012
Entered Medline: 20001005

AB Even though glycosylphosphatidylinositol (GPI)-anchored proteins lack direct structural contact with the intracellular space, these ubiquitously expressed surface receptors activate signaling cascades and endocytosis when **crosslinked** by extracellular ligands. Such properties may be due to their association with membrane microdomains composed of glycosphingolipids, cholesterol and some signaling proteins. In this study, we hypothesize that GPI proteins may be required for **crosslinking**-mediated endocytosis of extracellular bioconjugates. To test this hypothesis, we first biotinylated the surface membranes of native K562 erythroleukemia cells versus K562 cells incapable of surface GPI protein expression. We then compared the entry of fluorescently labeled **avidin** or DNA condensed on **polyethylenimine**-**avidin** bioconjugates into the two biotinylated cell populations. Using fluorescence microscopy, nearly 100% efficiency of fluorescent **avidin** endocytosis was demonstrated in both cell types over a 24 h period. Surprisingly, plasmid DNA transfer was slightly more efficient among the biotinylated GPI-negative cells as measured by the expression of green fluorescence protein. Our findings that GPI proteins are not required for the endocytosis of **avidin** bioconjugates into biotinylated cells suggest that endocytosis associated with general membrane **crosslinking** may be due to overall reorganization of the membrane domains rather than GPI protein-specific interactions.

L13 ANSWER 3 OF 6 MEDLINE on STN DUPLICATE 2
ACCESSION NUMBER: 2000031555 MEDLINE
DOCUMENT NUMBER: PubMed ID: 10563774
TITLE: Surface membrane biotinylation efficiently mediates the endocytosis of avidin bioconjugates into nucleated cells.

AUTHOR: Wojda U; Goldsmith P; Miller J L
CORPORATE SOURCE: Laboratory of Chemical Biology, National Institute of Diabetes and Digestive and Kidney Diseases and Metabolic Disease Branch, National Institutes of Health, Bethesda, Maryland 20892, USA.

SOURCE: Bioconjugate chemistry, (1999 Nov-Dec) 10 (6) 1044-50.
Journal code: 9010319. ISSN: 1043-1802.

PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 200001
ENTRY DATE: Entered STN: 20000114
Last Updated on STN: 20000114
Entered Medline: 20000106

AB Here we demonstrate that biotin **covalently** attached to cell surface obligates existing receptors to endocytose **avidin** bioconjugates into nucleated cells. Incubation of fluorescein-labeled **avidin** with biotinylated cell lines resulted in uniform and rapid surface attachment and endocytosis compared with no detectable association of the **avidin**-conjugated dye with unbiotinylated cells. Uptake

was detected within minutes with efficiencies approaching 100% in cell lines and freshly obtained peripheral blood mononuclear cells. After 24 h, **avidin** was barely detectable on the surface of the nucleated cells. In marked contrast, fluorescent **avidin** remained exclusively on the external membrane of erythrocytes after 24 h. To investigate biotin-mediated endocytosis for the delivery of DNA, we prepared **polyethylenimine-avidin** (**PEI-avidin**) conjugates. Surface biotinylation significantly increased the transfection efficiencies of **PEI-avidin** condensed plasmid DNA coding green fluorescent protein (GFP) to the level of transferrin-receptor targeted gene delivery (15-20% GFP positive cells in culture after 48 h). The increase in transfection efficiency was blocked by the addition of free **avidin** or biotin to the culture medium. Biotin **covalently** bound to cell surface membrane proteins efficiently mediates the entry of **avidin** bioconjugates into nucleated cells.

L13 ANSWER 4 OF 6 MEDLINE on STN DUPLICATE 3
ACCESSION NUMBER: 1999433860 MEDLINE
DOCUMENT NUMBER: PubMed ID: 10502353
TITLE: Polyion complex micelles with protein-modified corona for receptor-mediated delivery of oligonucleotides into cells.
AUTHOR: Vinogradov S; Batrakova E; Li S; Kabanov A
CORPORATE SOURCE: Department of Pharmaceutical Sciences, College of Pharmacy, 986025 Nebraska Medical Center Omaha, Nebraska 68198-6025, USA.
SOURCE: Bioconjugate chemistry, (1999 Sep-Oct) 10 (5) 851-60. Journal code: 9010319. ISSN: 1043-1802.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199911
ENTRY DATE: Entered STN: 20000111
Last Updated on STN: 20000111
Entered Medline: 19991119

AB Graft-copolymers, containing poly(ethylene glycol) (PEG) and polyethyleneimine (**PEI**) chains have been proposed as carriers for delivery of phosphorothioate oligonucleotides (SODNs). Complexes of such copolymers with SODN self-assemble into particles having a core of neutralized **PEI** and SODN and a corona of PEG. Transferrin molecules are attached to the PEG corona using **avidin**/biotin construct. For this purpose, biotin moieties are **covalently linked** to the free ends of the PEG chains in the PEG-g-**PEI** copolymer. SODNs are reacted with mixtures of biotinylated and biotin-free PEG-g-**PEI** copolymers of various compositions to adjust the number of the biotin moieties in the complex. Resulting complexes have small size (ca. 40 nm) and do not aggregate in aqueous solutions for at least several days. To attach transferrin, they are supplemented first with **avidin** and then with biotin-transferrin conjugate. This increases the effective diameter of the particles to ca. 75-103 nm, depending on the composition of the complex. Cellular accumulation and fluorescence microscopy studies characterize the effects of these modifications on interaction of fluorescently labeled SODNs with KBv cell monolayers. The data suggest significant enhancement of SODN association with cells resulting from modification of the complex with transferrin. SODN complimentary to the site 546-565 of human mdr 1-mRNA was used to inhibit expression of the drug efflux transporter, P-glycoprotein (P-gp), in multiple drug resistant (MDR) cancer cells (KBv, MCF-7 ADR). Accumulation of a P-gp specific probe, rhodamine 123, in the cell monolayers is used to characterize the effects on P-gp efflux system following the treatment of the cells with antisense SODN or its complexes.

This study suggests that antisense SODN incorporated in the complexes retain the ability to inhibit P-gp efflux system, while complexes of the randomized control SODN are inactive. Therefore, the antisense SODN is released from the complex and interacts with its intracellular target upon interaction of the complexes with the cells. Furthermore, modification of the complexes with transferrin leads to a significant increase of the effects of the antisense SODN on the P-gp efflux system in the cells. Overall, this study suggests that polyion complex micelles with protein-modified corona are promising tools for the delivery of antisense SODN.

L13 ANSWER 5 OF 6 MEDLINE on STN DUPLICATE 4
ACCESSION NUMBER: 97479353 MEDLINE
DOCUMENT NUMBER: PubMed ID: 9338005
TITLE: Polyethylenimine (PEI) is a simple, inexpensive and effective reagent for condensing and linking plasmid DNA to adenovirus for gene delivery.
AUTHOR: Baker A; Saltik M; Lehrmann H; Killisch I; Mautner V; Lamm G; Christofori G; Cotten M
CORPORATE SOURCE: Institute for Molecular Pathology, Vienna, Austria.
SOURCE: Gene therapy, (1997 Aug) 4 (8) 773-82.
Journal code: 9421525. ISSN: 0969-7128.
PUB. COUNTRY: ENGLAND: United Kingdom
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199711
ENTRY DATE: Entered STN: 19971224
Last Updated on STN: 19971224
Entered Medline: 19971119

AB A simple and inexpensive method of condensing and **linking** plasmid DNA to carrier adenovirus particles is described. The synthetic polycation **polyethylenimine** is used to condense plasmid DNA into positively charged 100 nm complexes. These **PEI**-DNA complexes are then bound to adenovirus particles through charge interactions with negative domains on the viral hexon. The resulting transfection complexes deliver plasmid DNA to cells by the adenovirus infectious route without interference from virus gene expression because psoralen-inactivated virus is employed. The **PEI**-DNA-adenovirus complexes display DNA delivery comparable to more sophisticated DNA virus complexes employing **streptavidin**/biotin **linkage**, but require no special reagents and are much easier to prepare.

L13 ANSWER 6 OF 6 CAPLUS COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 1989:420488 CAPLUS
DOCUMENT NUMBER: 111:20488
TITLE: Immobilization of polynucleotides on a plastic support with PEI for hybridization assays
INVENTOR(S): Pedley, Stephen
PATENT ASSIGNEE(S): National Research Development Corp., UK
SOURCE: Brit. UK Pat. Appl., 20 pp.
CODEN: BAXXDU
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
GB 2197720	A1	19880525	GB 1986-27703	19861120

PRIORITY APPLN. INFO.: GB 1986-27703 19861120

AB A polynucleotide is immobilized to a plastic support (e.g. a microtiter

plate) via a branched **PEI** which bonds to the plate and can be **crosslinked** to the polynucleotide with glutaraldehyde. A polynucleotide analyte can be assayed by binding it to the plate, probing it with a labeled complementary polynucleotide under hybridization conditions, and detecting the presence or amount of label. PVC microtiter plates were coated with **PEI** (10 mg/mL in 5 mM Na phosphate buffer, pH 6.8). The cDNA to foot-and-mouth disease virus RNA was denatured in boiling water, cooled rapidly, and added (50 μL) to the wells, followed by addition of 6 μL glutaraldehyde solution (5 g/100 mL in phosphate buffer) and incubation at 37° for 30 min. The immobilized DNA was hybridized with single-stranded, biotin-labeled (by nick-translation) viral cDNA 50-200 nucleotides long and detected by use of a **streptavidin**-biotinylatd peroxidase conjugate and o-phenylenediamine.

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=> D HIS

     (FILE 'HOME' ENTERED AT 09:32:58 ON 02 MAR 2004)

     FILE 'MEDLINE, CAPLUS, BIOSIS, EMBASE' ENTERED AT 09:33:21 ON 02 MAR 2004
L1       271771 S (WOJDA, ?)/IN,AU OR (MILLER, ?)/IN,AU OR (GOLDSMITH, ?)/IN,AU
L2            4 S (WOJDA, ?)/IN,AU AND (MILLER, ?)/IN,AU AND (GOLDSMITH, ?)/IN,
L3            2 DUPLICATE REMOVE L2 (2 DUPLICATES REMOVED)
L4            0 S L1 AND SULFO-NHS-LC-BIOTIN
L5            0 S L1 AND "SULFO-NHS-LC--LC-BIOTIN"
L6            1 S  "SULFO-NHS-LC--LC-BIOTIN"
L7           32 S SULFO-NHS-LC-BIOTIN
L8           13 DUPLICATE REMOVE L7 (19 DUPLICATES REMOVED)
L9        11610 S POLYETHYLENIMINE OR PEI
L10          28 S L9 (S) (AVIDIN OR STREPTAVIDIN)
L11          36 S L9 (P) (AVIDIN OR STREPTAVIDIN)
L12          16 S L11 (P) (COVALENT? OR LINK? OR CROSSLINK?)
L13           6 DUPLICATE REMOVE L12 (10 DUPLICATES REMOVED)

=> S L12 NOT L13
L14          10 L12 NOT L13

=> S L11 NOT L12
L15          20 L11 NOT L12

=> DUPLICATE REMOVE L15
DUPLICATE PREFERENCE IS 'MEDLINE, CAPLUS, BIOSIS, EMBASE'
KEEP DUPLICATES FROM MORE THAN ONE FILE?  Y/(N):N
PROCESSING COMPLETED FOR L15
L16          11 DUPLICATE REMOVE L15 (9 DUPLICATES REMOVED)

=> D IBIB AB L16 1-11

L16  ANSWER 1 OF 11  CAPLUS  COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER:         2003:167694  CAPLUS
DOCUMENT NUMBER:          138:360905
TITLE:                    Electronic detection of specific protein binding using
                          nanotube FET devices
AUTHOR(S):                Star, Alexander; Gabriel, Jean-Christophe P.; Bradley,
                          Keith; Gruener, George
CORPORATE SOURCE:         Nanomix Inc., Emeryville, CA, 94608, USA
SOURCE:                   Nano Letters (2003), 3(4), 459-463
                          CODEN: NALEFD; ISSN: 1530-6984
PUBLISHER:                American Chemical Society
DOCUMENT TYPE:            Journal
LANGUAGE:                 English
```

AB The authors use nanoscale field effect transistor devices with C nanotubes as the conducting channel to detect protein binding. A **PEI**/PEG polymer coating layer has been employed to avoid non-specific binding, with attachment of biotin to the layer for specific mol. recognition. Biotin-**streptavidin** binding has been detected by changes in the device characteristic. Non-specific binding was observed in devices without the polymer coating, while no binding was found for polymer-coated but not biotinylated devices. **Streptavidin**, in which the biotin-binding sites were blocked by reaction with excess biotin, produced essentially no change in device characteristic of the biotinylated polymer-coated devices.

REFERENCE COUNT: 27 THERE ARE 27 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L16 ANSWER 2 OF 11 CAPLUS COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 2002:10674 CAPLUS
DOCUMENT NUMBER: 136:65207
TITLE: Method for transfecting cells using a magnetic field
INVENTOR(S): Plank, Christian; Bergemann, Christian
PATENT ASSIGNEE(S): Germany
SOURCE: PCT Int. Appl., 125 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 2
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2002000870	A2	20020103	WO 2001-EP7261	20010626
WO 2002000870	A3	20020704		
WO 2002000870	C2	20020919		

W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM
RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG

US 2002086842	A1	20020704	US 2001-895019	20010626
EP 1297169	A2	20030402	EP 2001-949436	20010626

R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO, MK, CY, AL, TR

PRIORITY APPLN. INFO.:

EP 2000-113083	A	20000626
US 2000-214286P	P	20000626
WO 2001-EP7261	W	20010626

AB Described is a method of cell transformation by using a complex comprising vector(s) and magnetic particle(s) in contact with the cell, and by applying a magnetic field. The methods for preparation of the complex is presented. Finally, pharmaceutical compns., uses in such complexes and a kit are described. The developed method is particularly useful when automation of high-throughput transfection is required for large scale screening processes.

L16 ANSWER 3 OF 11 CAPLUS COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 2002:676922 CAPLUS
DOCUMENT NUMBER: 139:57746
TITLE: Targeted oligonucleotide delivery in human lymphoma cell lines using a polyethyleneimine based immunopolyplex

AUTHOR(S): Guillem, Vicent M.; Tormo, Mar; Moret, Ines; Benet, Isabel; Garcia-Conde, Javier; Crespo, Antonio; Alino, Salvador F.
CORPORATE SOURCE: Facultat de Medicina, Hospital Clinic Universitari, Servei d' Hematologia i Oncologia, Universitat de Valencia, Valencia, 46010, Spain
SOURCE: Journal of Controlled Release (2002), 83(1), 133-146
CODEN: JCREEC; ISSN: 0168-3659
PUBLISHER: Elsevier Science Ltd.
DOCUMENT TYPE: Journal
LANGUAGE: English

AB The efficacy of antisense gene therapy depends on efficient delivery of oligonucleotides into targeted cells. Although polyethyleneimine based polyplexes have been reported as good transfection reagents, they are inefficient in lymphoid cell transfection. We report the construction of an immunopolyplex, a targeted nonviral vector based on a polyplex backbone and its application for oligonucleotide transfer on human lymphoma cell lines. The salient characteristic of immunopolyplex lies in the possibility of easily replacing the targeting element (antibody), leaving the polyplex backbone intact. Furthermore, a study was made of the influence of endocytosis inhibitors on immunopolyplex activity. The capacity of the immunopolyplex for oligonucleotide transfer was studied in vitro using FITC-labeled oligonucleotides as fluorescent reporters, an anti-CD3 antibody as targeting element, and a CD3-pos. cell line (Jurkat) as a target cell line, in the absence and presence of endocytosis inhibitors. A CD3-neg. Jurkat-derived mutant cell line (J.RT3-T3.5) was used as control. A nine-fold increase in fluorescence in the CD3-pos. above that in the CD3-neg. cell line was observed, indicating that oligonucleotide transfer is mainly specific. Low fluorescence values were obtained in the presence of endocytosis inhibitor or with untargeted polyplexes. We conclude that the immunopolyplex is a good nonviral vector for specific oligonucleotide delivery. Abolition of immunopolyplex activity in the presence of endocytosis inhibitor suggests that targeted oligonucleotide transfer occurs through an endocytic pathway.

REFERENCE COUNT: 45 THERE ARE 45 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L16 ANSWER 4 OF 11 MEDLINE on STN DUPLICATE 1
ACCESSION NUMBER: 2002462074 MEDLINE
DOCUMENT NUMBER: PubMed ID: 12220843
TITLE: A receptor-mediated gene delivery system using streptavidin and biotin-derivatized, pegylated epidermal growth factor.
AUTHOR: Lee Haeshin; Kim Tae Hyoung; Park Tae Gwan
CORPORATE SOURCE: Department of Biological Sciences, Korea Advanced Institute of Science and Technology, Taejon 305-701, South Korea.
SOURCE: Journal of controlled release : official journal of the Controlled Release Society, (2002 Sep 18) 83 (1) 109-19.
Journal code: 8607908. ISSN: 0168-3659.
PUB. COUNTRY: Netherlands
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 200212
ENTRY DATE: Entered STN: 20020911
Last Updated on STN: 20021220
Entered Medline: 20021219

AB An efficient receptor-mediated non-viral gene delivery formulation based on mono-pegylated recombinant human epidermal growth factor (EGF) was developed using a **streptavidin**-biotin system.
Biotin-derivatized and mono-pegylated EGF was prepared by conjugating a biotin-PEG-NHS derivative to EGF and purified through a chromatographic method. Luciferase plasmid DNA and **polyethylenimine** (

PEI) were complexed to form positively charged nanoparticles on which negatively charged **streptavidin** was first coated and then biotin-PEG-EGF conjugate was immobilized via **streptavidin**-biotin interaction. The EGF-PEG-biotin-**streptavidin**-**PEI**-DNA complexes were characterized in terms of their effective diameter and surface zeta (zeta)-potential value under various formulation conditions. The formulated complexes exhibited high transfection efficiency (approximately 10(8) in luciferase activity) with no inter-particle aggregation. This was attributed to enhanced cellular uptake of the resultant complexes via receptor-mediated endocytosis. Furthermore, in the presence of serum proteins, a slight decrease in transfection efficiency was observed due to the presence of PEG chains on the surface. Copyright 2002 Elsevier Science B.V.

L16 ANSWER 5 OF 11 MEDLINE on STN DUPLICATE 2

ACCESSION NUMBER:	2000221506 MEDLINE
DOCUMENT NUMBER:	PubMed ID: 10756333
TITLE:	Targeted transfer of **polyethylenimine**-**avidin**-DNA bioconjugates to hematopoietic cells using biotinylated monoclonal antibodies.
AUTHOR:	Wojda U; Miller J L
CORPORATE SOURCE:	Laboratory of Chemical Biology, National Institute of Diabetes and Digestive and Kidney Diseases, National Institutes of Health, 9000 Rockville Pike, Bethesda, Maryland 20892, USA.
SOURCE:	Journal of pharmaceutical sciences, (2000 May) 89 (5) 674-81. Journal code: 2985195R. ISSN: 0022-3549.
PUB. COUNTRY:	United States
DOCUMENT TYPE:	Journal; Article; (JOURNAL ARTICLE)
LANGUAGE:	English
FILE SEGMENT:	Priority Journals
ENTRY MONTH:	200007
ENTRY DATE:	Entered STN: 20000811 Last Updated on STN: 20000811 Entered Medline: 20000728

AB Here we examine whether attachment of biotinylated antibodies to proteins on the cell surface increases the transfection efficiency of **polyethylenimine**-**avidin**-DNA bioconjugate gene transfer. Preliminary experiments were performed to compare **avidin** endocytosis into cells incubated with biotinylated antibodies. Antibody biotinylation resulted in the endocytosis of **avidin**-FITC into nearly 100% of cells compared with no detectable binding or entry into unbiotinylated cells. Gene transfer was accomplished with **avidin** conjugated to **polyethylenimine** (**PEI**) at a molar ratio of 4:1 (PA4). Plasmid DNA encoding the green fluorescent protein (GFP) gene was condensed on the PA4, and transfection efficiencies were measured by flow cytometry as the percentage of cells that fluoresced at levels greater than two standard deviations above the negative control. Gene transfer efficiencies were compared among K562, HEL, and Jurkat leukemia cell lines. Control transfections with DNA alone or untargeted **PEI**-DNA resulted in </=2% GFP positive cells. Targeting **PEI**-**avidin**-DNA to antibody biotinylated cells increased transfection efficiency several fold over untargeted **PEI**. For each cell type, the increase in transfection efficiency was not significantly different among four biotinylated antibodies tested (antiCD55, antiCD59, antiCD71, and antiCD98). These data suggest biotinylated antibodies may be useful for targeting **polyethylenimine**-**avidin** mediated gene transfer.
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L16 ANSWER 6 OF 11 CAPLUS COPYRIGHT 2004 ACS on STN

ACCESSION NUMBER: 1998:772033 CAPLUS
DOCUMENT NUMBER: 130:121813
TITLE: Layer-by-Layer Construction of Multilayer Thin Films Composed of Avidin and Biotin-Labeled Poly(amine)s
AUTHOR(S): Anzai, Jun-ichi; Kobayashi, Yuka; Nakamura, Nobuyuki; Nishimura, Masahiro; Hoshi, Tomonori
CORPORATE SOURCE: Faculty of Pharmaceutical Sciences, Tohoku University, Aramaki Aoba-ku Sendai, 980-8578, Japan
SOURCE: Langmuir (1999), 15(1), 221-226
CODEN: LANGD5; ISSN: 0743-7463
PUBLISHER: American Chemical Society
DOCUMENT TYPE: Journal
LANGUAGE: English

AB Multilayer thin films composed of a pos. charged protein and cationic polymer layers can be prepared on a solid surface against their electrostatic force of repulsion, by depositing **avidin** and biotin-labeled polymers [poly(ethyleneimine) (**PEI**), poly(allylamine) (PAA), or poly(amidoamine) (PAMAM) dendrimer] alternately and repeatedly. In contrast, the thin film does not form when unmodified polymers are used, confirming that the strong binding between **avidin** and biotin is responsible for the formation of the multilayer films. The loading of **avidin** in each layer of the multilayer films depends significantly on the mol. geometry of the polymers. The **avidin**/PAMAM dendrimer films are composed of a monomol. layer of **avidin** whereas the **avidin** multilayers are formed in each layer of the PAA- and **PEI**-based thin films. The results are rationalized by the different mol. structures of the polymers; PAMAM dendrimer assumes a globular shape, as compared with the linear and highly branched polymer chains of PAA and **PEI**, resp.

REFERENCE COUNT: 38 THERE ARE 38 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L16 ANSWER 7 OF 11 CAPLUS COPYRIGHT 2004 ACS on STN DUPLICATE 3
ACCESSION NUMBER: 1999:653450 CAPLUS
DOCUMENT NUMBER: 132:185291
TITLE: Studies on the construction of protein/DNA conjugates for targeted transfection of cells using receptor-mediated endocytosis
AUTHOR(S): Hawtrey, A. O.; Ariatti, M.
CORPORATE SOURCE: Department of Pharmacology, Medical School, University of Stellenbosch, Tygerberg, 7505, S. Afr.
SOURCE: S.T.P. Pharma Sciences (1999), 9(1), 35-45
CODEN: STSSE5; ISSN: 1157-1489
PUBLISHER: Editions de Sante
DOCUMENT TYPE: Journal; General Review
LANGUAGE: English

AB A review with 60 refs. Delivery of genes to cells via receptor-mediated endocytosis requires attachment of a specific receptor ligand to the gene (DNA) of interest. Methods for this are discussed from both historical and theor. aspects, while exptl. work on the ligand-receptor approach is also outlined. Use of poly-L-lysine and **polyethylenimine** for attachment and condensation of the DNA component as well as for binding chemical modified transferrin and asialo α1 acid-glycoprotein receptor ligands is discussed. Biotinylated transferrin and biotinylated polylysine conjugates of **avidin**/**streptavidin** with attached DNA also form interesting transfecting complexes. The effect of coupled polyethyleneglycol on the transfecting ability of these macromol. assemblies is examined as well as various factors influencing delivery of DNA to a cell and its final destination in the nucleus.

REFERENCE COUNT: 60 THERE ARE 60 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L16 ANSWER 8 OF 11 BIOSIS COPYRIGHT 2004 BIOLOGICAL ABSTRACTS INC. on STN
ACCESSION NUMBER: 1999:111564 BIOSIS
DOCUMENT NUMBER: PREV199900111564
TITLE: Gene delivery to biotinylated hematopoietic cells using **avidin-polyethylenimine** bioconjugates.
AUTHOR(S): Wojda, U.; Goldsmith, P.; Miller, J. L.
CORPORATE SOURCE: Lab. Chem. Biol. Metabol. Dis. Branch, Natl. Inst Diabetes Digestive Kidney Dis., NIH, Bethesda, MD, USA
SOURCE: Blood, (Nov. 15, 1998) Vol. 92, No. 10 SUPPL. 1 PART 1-2, pp. 382B. print.
Meeting Info.: 40th Annual Meeting of the American Society of Hematology. Miami Beach, Florida, USA. December 4-8, 1998. The American Society of Heamatology.
CODEN: BLOOAW. ISSN: 0006-4971.
DOCUMENT TYPE: Conference; (Meeting)
Conference; Abstract; (Meeting Abstract)
LANGUAGE: English
ENTRY DATE: Entered STN: 12 Mar 1999
Last Updated on STN: 12 Mar 1999

L16 ANSWER 9 OF 11 CAPLUS COPYRIGHT 2004 ACS on STN DUPLICATE 4
ACCESSION NUMBER: 1992:37199 CAPLUS
DOCUMENT NUMBER: 116:37199
TITLE: A novel approach to nonradioactive hybridization assay of nucleic acids using stained latex particles
AUTHOR(S): Vener, T. I.; Turchinskii, M. F.; Knorre, V. D.; Lukin, Yu. V.; Shcherbo, S. N.; Zubov, V. P.; Sverdlov, E. D.
CORPORATE SOURCE: M. M. Shemyakin Inst. Bioorg. Chem., Moscow, 117871, USSR
SOURCE: Analytical Biochemistry (1991), 198(2), 308-11
CODEN: ANBCA2; ISSN: 0003-2697
DOCUMENT TYPE: Journal
LANGUAGE: English
AB Described is a sensitive latex hybridization assay (LHA) applied to indirect detection of biotinylated nucleic acid hybrids immobilized on a synthetic membrane. The biotinylated hybrids were visualized by means of polyacrolein latex particles containing the fluorescent dye pyronine G and coated with **streptavidin**; 1.6 and 0.3 pg of λ-phage DNA was detected by dot-blot hybridizations on nylon membrane and **polyethylenimine**-cellophane, resp. The assay sensitivity was increased by 3 orders of magnitude over that with fluorescently labeled probes due to encapsulation of the fluorescent dye in polymer particles. LHA is simple (single-stage detection procedure), fast, and more sensitive than any of the other nonradioactive hybridization methods.

L16 ANSWER 10 OF 11 BIOSIS COPYRIGHT 2004 BIOLOGICAL ABSTRACTS INC. on STN
ACCESSION NUMBER: 1990:412349 BIOSIS
DOCUMENT NUMBER: PREV199090073150; BA90:73150
TITLE: FURTHER OBSERVATIONS ON THE STRUCTURE AND COMPOSITION OF THE CELL-COAT OF DUNALIELLA-BIOCULATA GREEN ALGA.
AUTHOR(S): CHARDARD R [Reprint author]
CORPORATE SOURCE: 89BIS AVENUE DU GENERAL DE GAULLE, 94160 ST-MANDE, FRANCE
SOURCE: Cryptogamie Algologie, (1990) Vol. 11, No. 2, pp. 137-152.
CODEN: CRALD9. ISSN: 0181-1568.
DOCUMENT TYPE: Article
FILE SEGMENT: BA
LANGUAGE: FRENCH
ENTRY DATE: Entered STN: 17 Sep 1990
Last Updated on STN: 17 Sep 1990
AB Further studies have been carried out on the ultrastructure and

composition of the cell-coat of Dunaliella bioculata Butcher by cytochemical techniques. The use of polyethyleneimine (**PEI**) of various molecular weights has confirmed the presence of negative charges on the cell-coat and illustrated their distribution. Ferritin labelled lectins have also been used. Thus, WGA [wheat germ agglutinins] coupled with biotin and detected by **avidin**-ferritin is seen to be fixed on the cell-coat suggesting the presence of N-acetyl glucosamine in glycoproteins. On other hand, Con A gives a negative response.

L16 ANSWER 11 OF 11 CAPLUS COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 1989:72076 CAPLUS
DOCUMENT NUMBER: 110:72076
TITLE: Time-resolved immunofluorescence assay using europium labels and polymeric complexon
AUTHOR(S): Krylova, S. M.; Sakharov, I. Yu.; Slinkin, M. A.; Savitskii, A. P.; Kibanov, L.; Torchilin, V. P.; Berezin, I. V.
CORPORATE SOURCE: Inst. Biochem., Moscow, USSR
SOURCE: Voprosy Meditsinskoi Khimii (1988), 34(5), 116-21
CODEN: VMDKAM; ISSN: 0042-8809
DOCUMENT TYPE: Journal
LANGUAGE: Russian

AB The proposed solid-phase immunofluorescence assay uses a new method of binding rare earth label Eu with antibodies. The stages include: adsorption of mouse Ig as antigens on polystyrene microspheres; immunol. binding reaction between antigens and biotinylated antibodies (rabbit Ig against mice IgG). Binding of biotinylated antibodies with **avidin** ; binding of **avidin** with complexon biotin-**polyethylenimine**-DTPA-Eu; desorption of Eu from the microspheres and its detection. Optimum concentration of **avidin** was 2-10 μg/mL and of biotinylated polymer containing Eu was 5 + 10-1-5 + 10-6M. The sensitivity of the method was 0.01 μg/mL. The sensitivity of EIA using biotin-peroxidase conjugates was an order less than that by this method.

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(FILE 'HOME' ENTERED AT 09:32:58 ON 02 MAR 2004)

FILE 'MEDLINE, CAPLUS, BIOSIS, EMBASE' ENTERED AT 09:33:21 ON 02 MAR 2004
L1 271771 S (WOJDA, ?)/IN,AU OR (MILLER, ?)/IN,AU OR (GOLDSMITH, ?)/IN,AU
L2 4 S (WOJDA, ?)/IN,AU AND (MILLER, ?)/IN,AU AND (GOLDSMITH, ?)/IN,
L3 2 DUPLICATE REMOVE L2 (2 DUPLICATES REMOVED)
L4 0 S L1 AND SULFO-NHS-LC-BIOTIN
L5 0 S L1 AND "SULFO-NHS-LC--LC-BIOTIN"
L6 1 S "SULFO-NHS-LC--LC-BIOTIN"
L7 32 S SULFO-NHS-LC-BIOTIN
L8 13 DUPLICATE REMOVE L7 (19 DUPLICATES REMOVED)
L9 11610 S POLYETHYLENIMINE OR PEI
L10 28 S L9 (S) (AVIDIN OR STREPTAVIDIN)
L11 36 S L9 (P) (AVIDIN OR STREPTAVIDIN)
L12 16 S L11 (P) (COVALENT? OR LINK? OR CROSSLINK?)
L13 6 DUPLICATE REMOVE L12 (10 DUPLICATES REMOVED)
L14 10 S L12 NOT L13
L15 20 S L11 NOT L12
L16 11 DUPLICATE REMOVE L15 (9 DUPLICATES REMOVED)

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